THE

AMERICAN JOURNAL

OF THE MEDICAL SCIENCES.

APRIL, 1889.

ON THE ETIOLOGY OF DIPHTHERIA.

AN EXPERIMENTAL STUDY.

BY T. MITCHELL PRUDDEN, M.D.,
DIRECTOR OF THE LABORATORY OF THE ALUMNI ASSOCIATION OF THE COLLEGE OF PHYSICIANS AND SURGEONS, NEW YORK.

PART I.

THE thoughtful student of diphtheria is early impressed, alike in his studies by the bedside, at the dissecting-table, and in the laboratory, by the indefinite conception which so widely prevails as to the nature and limitations of the disease. The difficulties with which this lack of precision invests the subject are especially felt when he is trying to get light upon its etiology. The standpoint from which these studies are undertaken can, perhaps, be best made clear by a brief statement of the views which the writer believes to be generally held in this country as to the general nature of diphtheria and its relations to diseases not infrequently confounded with it.

Diphtheria, in its more pronounced forms, is an acute infectious disease, presenting in most cases a more or less characteristic local lesion in some of the external or internal surfaces of the body. This local lesion is most frequent in the pharynx and upper air-passages, but may occur on any mucous membrane, or on the skin, or on the surfaces of wounds.

This local inflammation may find expression in a variety of ways. There may be a simple redness of the affected surfaces, which passes away leaving no trace. There may be a more or less intense catarrhal inflammation with much or little exudation, and nothing else. There may be a limited or extended superficial or deep necrosis of the mucous membrane, a form of necrosis called coagulation-necrosis, in which the nuclei of the cells disappear, and the cell bodies are converted into a more or less homogeneous or granular mass somewhat resembling fibrin, so that the affected surfaces appear to be covered with a whitish or grayish pellicle or membrane. There may be, in connection with or without this necrotic pellicle, a fibrinous exudate intermingled with leucocytes, fragments of epithelial cells, red-blood cells, and granular matter.

The pellicle, or false membrane, pseudo-membrane as it is called, may be loosely attached to the affected surfaces, or so firmly adherent as to seem to form a part of the underlying tissues. Associated with all these forms of the local lesion of diphtheria are varying, but usually large, numbers of dead and living microörganisms. Disintegration and softening or exfoliation of the false membrane, with or without loss of substance of the underlying tissue, ulceration, phlegmon, œdema, and abscess, represent occasional progressive phases or complications of the lesion. The adjacent lymph-glands are apt to be swollen and congested.

It is not necessary to describe here in more detail the minute structure of the false membrane in diphtheria, nor the various minor modifications which it presents. Nor is it necessary to do more than call attention to the fact that, although the occurrence of a false membrane is very frequent in the disease and of great diagnostic importance, there are cases of genuine diphtheria in which no false membrane is formed.

It is, however, highly important to remember that, so far as its anatomical characters are concerned, a false membrane very similar to that above described is by no means limited in its occurrence to diphtheria. A pseudo-membrane may be induced experimentally in animals, as it would seem, by any agency which will deprive mucous membranes, such as that of the trachea, of their epithelial covering, and fulfil the conditions of fibrin-generation through the induction of emigration of leucocytes. It may be formed under the influence of a great variety of local irritants, such as steam, cantharides, corrosive sublimate, arsenic, sulphuric acid, ammonia, etc. It may also be formed as Huebner (1) has shown, by such a local temporary mechanical interference with the circulation in the affected part as will impair the integrity of its vascular endothelium.

Under all of these conditions, then, a false membrane may be formed essentially similar in anatomical character, so far as we yet know, save, perhaps, for the microörganisms which it contains, to some of the forms which are frequent in diphtheria. This false membrane is often called a croupous membrane, and the form of inflammation which it accompanies a croupous inflammation. It is also, unfortunately, termed a diphtheritic membrane alike whether it occurs in diphtheria or in inflammation due to a variety of other local causes.

The attempt to establish an anatomical distinction between those forms, especially of pharyngeal and laryngeal inflammation with the formation of a false membrane, which, on the one hand, seemed to be of a purely local character, and, on the other, were associated with evidences of systemic infection; the attempt, in other words, to establish a morphological criterion for the differentiation of what is known as croup or simple pseudo-membranous inflammation of the larynx and of the pharynx from a more serious form of disease called diphtheria, was a legitimate attempt to meet an urgent demand for precision in the interests both of treatment and prophylaxis. Especially under Virchow's teaching, it was at one time generally believed that, if the pseudo-membrane had evidently encroached upon the structure of the underlying parts, so that its removal involved a loss of substance, its formation was due to those agencies which induced the disease known as diphtheria. If, on the other hand, the pellicle was superficial, it was the result of a simple local croupous inflammation, and might be due to a variety of causes. When, however, it became evident, as it soon did, that in very many cases in which a more or less extensive pharyngeal or laryngeal inflammation with a pseudo-membrane, was associated with grave systemic reaction, the false membrane was apparently entirely superficial, and could be stripped off without injury to the underlying tissue; when, on the other hand, a similar local inflammation developed with no evidences whatsoever of systemic infection, and with no tendency to the spread of the disease from one individual to another, and still it was found that there might be a close adherence, even a genuine infiltration of the underlying tissue with the false membrane: it became evident that the deep or superficial character of the lesion was a distinction between simple pseudo-membranous pharyngitis or laryngitis and diphtheria which, in fact, did not distinguish.

In the reaction which followed the loss of faith in this anatomical distinction between simple croupous and diphtheritic inflammation, the often repeated observation that cases running the ordinary course of a simple local croupous inflammation with no systemic reaction, might apparently serve as a starting-point of widespread epidemics of genuine diphtheria, led many to take the ground that every inflammation with the formation of a pseudo-membrane which was not evidently traumatic —*i. e.*, due either to chemical or mechanical injury—should be regarded as diphtheria. In other words, the view seems generally to have become prevalent that it is at least safe and wise to consider those forms of disease commonly known as simple croup or pseudo-membranous laryngitis and pseudo-membranous pharyngitis as all local lesions of diphtheria, but diphtheria in which the symptoms of general infection are sometimes well defined and sometimes very little marked or absent altogether.

Whether the material which induces the inflammation in diphtheria sometimes reaches the point of local expression through the bloodvessels, or always from the surface in the affected region, it is not possible now absolutely to say, but both clinical and experimental evidences speak strongly in favor of the latter view.

The observations, largely clinical, but in part anatomical, which have led to the general belief that diphtheria is an acute infectious disease are too well known to need recapitulation here. If it be an acute infectious disease, the great advances in our knowledge of other diseases of this class would justify us in the conjecture that it is caused by bacteria and by some one or more particular species. Should such species, proved to stand in an etiological relationship to the disease, be ultimately discovered in the local lesions, most of the difficulties of definition which now beset our path would be largely removed. In other words, it is the etiological factor in diphtheria which we need to know in order to make those exact distinctions between it and other diseases with a similar local lesion, which the exigencies both of prophylaxis and treatment demand. When we know what causes diphtheria, then, and not till then, shall we know what diphtheria really is and how to define it. In the meantime we have to search for the etiological factor in the disease, without a more precise characterization of it than is furnished by the clinical observations in each particular case.

A large number of the older observers, in the use of the comparatively crude methods formerly in vogue, found bacteria of various forms in the membrane, and most of these seemed to belong to the group of micrococci, although bacilli are noted. But we have long since learned that the morphological examination does not suffice for the determination of a bacterial species. We must cultivate them in pure form to learn the much more definite biological features which characterize them. Not less indefinite were the results of the early attempts to reproduce the disease in animals. For the most part, the inoculation of animals, either subcutaneously or on the mucous membrane or directly into the blood, was practised with fragments of the false membrane, or with cultures from these, which, owing to the lack of proper methods, were never pure cultures of a single species, but a mixture of various forms. Under these conditions, sometimes in the animal a false membrane, or what passed for such, was produced; sometimes fatal septicæmia or local inflammatory lesions; sometimes all of these together, and sometimes none of them. But we should not forget that neither a false membrane nor septicæmia nor a simple local inflammation, nor even the simultaneous occurrence of these necessarily constitutes diphtheria.

We now know that there are many forms of bacteria which, when introduced into the bodies of animals, produce serious disturbance, marked disease, and even death. We know that some of the bacteria which are frequently present in the healthy human body, or its excretions, may, when introduced into the tissues or blood of animals, induce disease and death. So that now-a-days, when animals are inoculated with such unknown mixtures of various kinds of germs as may be in a diphtheritic membrane and die, our interpretation of the results is much more guarded and our conclusions much less sweeping than was formerly the case.

For our present purpose, then, we may pass without detailed review the long list of patient researches on the etiology of diphtheria which preceded the formulation of the new methods of bacterial study by Koch; not because they are without value or importance, but because the vantage-ground of the modern observer is on a so much higher plane of exactness and precision.[1] It implies no lack of respect for the conscientious workers of an older period of science, that at the advent of a new and revolutionizing mode of research the earlier results should for the moment be set aside until in the light of new and rapidly accumulating facts their significance can be justly estimated.

HISTORICAL SUMMARY.

The first exact study of the etiology of diphtheria by the use of the modern culture methods was by Loeffler, in 1883 (2). At this time, after a careful *résumé* of the work which had gone before, this observer came to the conclusion that no reliable data were at hand for fixing upon any particular species of bacteria as the cause of diphtheria; nor had any of the variously modified inoculations of animals, either with the crude membrane of diphtheria or the impure cultures from it, ever induced what could fairly be regarded as real diphtheria.

It will suffice for our purpose to give an outline of Loeffler's results. Two species of bacteria were found by him, sometimes alone, sometimes together, in so many cases as to render them of apparent significance. These were: 1, a chain coccus or streptococcus; and, 2, a slender bacillus.

So far as the chain coccus is concerned, although he apparently found it present by the morphological examination, in a considerable number of cases—as well in the blood and viscera as in the false membrane—he isolated and identified it by cultures in only five cases; from the tonsils in three of them; from the heart, liver, and spleen in two; from the kidney in one. Not only the biological characters of the chain coccus, but its general effects upon inoculated animals would seem to indicate its close relationship, if not its identity, with the streptococcus pyogenes and streptococcus erysipelatos. Its local effects upon inoculated mucous membrane did not at all resemble the characteristic common local lesions of diphtheria. Loeffler is disposed to consider this species, in most cases at least, as of secondary importance—though perhaps inducing complications; but his reasons for this view do not seem to be very precise or convincing.

---

[1] For a *résumé* of older researches on the etiology of diphtheria see Loeffler's studies, Bibliography, No. 2.

The bacilli of Loeffler are about as long on the average as the bacillus tuberculosis, and about twice as thick. They are frequently a little thickened or knobbed at the end. They stain readily with alkaline methylene-blue solutions. They are immobile, and do not grow at the ordinary temperature of the room. On blood-serum or on a mixture of blood-serum with beef-tea, peptone, grape-sugar, and salt, they grow readily at 37° C. (98.6° F.). They grow, also, as has since been shown, on nutrient agar, to which ten per cent. of glycerine has been added. These bacilli were obtained in pure culture from the false membranes in six cases of diphtheria. In one of these cases the streptococcus was also found. In four of them the material examined was the membrane simply, taken from children during life. By a simple morphological examination of seven other cases he found bacilli which appeared to be similar to those which he had cultivated. In the cases, however, which were examined both morphologically and by cultures, the bacilli do not seem to have been more frequently present than were the streptococci.

Cultures of the bacilli injected subcutaneously into guinea-pigs killed the animals within two to three days, producing a whitish or hemorrhagic exudate at the seat of inoculation and widespread œdema of the adjacent subcutaneous tissue. The internal organs were free from the inoculated bacilli, which were present only at the seat of inoculation. Introduced into the mucous membranes of rabbits, guinea-pigs, chickens, or pigeons, a fibrinous pellicle was frequently formed over the inoculated surface. Sometimes there were considerable general œdema and pleural exudation. On the other hand, the bacilli were not found in all typical cases of human diphtheria. They were absent in more than one-half of those examined morphologically, and in four out of ten in which cultures were made. The bacilli were not present in the fibrinous pellicles induced on animal mucous membrane either in numbers or situations corresponding to the human local lesion. They were often absent altogether. They did not seem to produce their effects on uninjured mucous membranes. They were found once in the mouth of a healthy child.

Thus while the researches of Loeffler led the way along lines which may be regarded in many respects as models for future workers, no positive conclusion was reached as to the cause of diphtheria. He concludes that while his bacilli are possibly the cause of diphtheria, the strict proof was not furnished by his investigations.

Loeffler is disposed to believe that there may be more than one kind of diphtheria; that the form of the disease in which the streptococci are present is characterized by a marked necrosis of the affected surface, while the pseudo-membrane is not prominently or extensively developed;

and, further, that the cocci enter the lymph channels and may find their way into various distant parts of the body, even blocking the smaller bloodvessels. That form of diphtheria, on the other hand, in which he finds the bacilli is characterized by an abundant pseudomembrane which may contain the bacilli in that part which borders on the original epithelial surface.

This view, that diphtheria might be caused by different species of microörganisms, had been already urged by Klebs (3), who had observed that the bacteria common in the false membrane in the cases which he had studied in Prague were of an entirely different form from those which he found in his later observations at Zurich. While in the former cases they were micrococci (microsporon diphtheriticum), in the latter they were bacilli apparently identical, morphologically, with those which the studies of Loeffler led him to regard as of much significance. The bacilli are now commonly known as the Klebs-Loeffler diphtheria bacilli.

In 1884, Emmerich (4) announced himself at the International Congress of Hygiene at the Hague as the discoverer of the real germ of diphtheria. He considers the disease in man and the pigeon as identical. His bacillus is short and stout, about twice as long as broad, is apt to occur in pairs. It grows luxuriantly in ordinary nutrient gelatine at 15° C. (59° F.), not fluidifying. On potato it grows vigorously, resembling the pneumococcus of Friedländer. He claims as the result of inoculations of pigeons, rabbits, and white mice, that he produces a disease which is usually fatal; that a pseudo-membrane forms in the trachea; and that the bacteria are found in the blood and in the internal organs, especially in the kidneys. Important as these results of Emmerich may seem to be, they have not been confirmed by others, and in several particulars there appear to be such possibilities of serious error in his technical procedures that it would seem unwise to attach a definite value to them until fuller detail or control research by others has given us more positive data. At a later period Emmerich (5), while still insisting upon the importance of his alleged diphtheria bacillus, calls attention to the frequent occurrence of streptococci in the diphtheritic membranes.

In 1886, Babes (6) reports the cultivation from six cases of pharyngeal diphtheria, from eight cases of croupous laryngitis, from three cases of diphtheria after measles, from one case of diphtheria after scarlatina, and from one case of diphtheria of the conjunctiva, of the bacillus of Loeffler among others. In these cases he found streptococci associated with Loeffler's bacillus. The details of his work are not given in this report nor is there any intimation that he tested the pathogenic properties of the isolated bacilli. In view of the incomplete character of this work and of the later discovery (see below) by Loeffler and v. Hoffmann, of a somewhat similar non-virulent bacillus, these results of Babes cannot be given much weight as evidence of the frequency of the occurrence or importance of Loeffler's bacillus in diphtheria.

In April, 1887, Loeffler (7) again made a communication upon the subject, in which he states that in ten cases in which fresh diphtheritic membranes were examined he found the bacilli in all. He also describes the occurrence in one of his cases of a bacillus very similar in its morphological and biological characters to those found in his earlier studies, but which was without effect upon animals. This he calls the "pseudo-diphtheritic bacillus."

Penzoldt (8), in 1887, as the result of a long series of somewhat rambling experiments found that he could not succeed in inducing typical diphtheria in rabbits, chickens, or pigeons by inoculation either with fresh diphtheritic membranes or with cultures from them. He sometimes obtained thick or thin, for the most part loose, pellicles over the inoculated surfaces, but these could be induced by a variety of bacteria which obviously have nothing to do with diphtheria. He gives but little detail as to the species which he isolated by culture. He emerges from his researches with a disposition to believe that diphtheria, in a clinical sense, may be due to a variety of causes.

Fränkel, in the same year, described a case, in an adult, of diphtheria of the fauces and larynx with secondary phlegmonous inflammation and pleurisy and pericarditis, from which he cultivated the streptococcus pyogenes (9). He describes another case, also an adult, of diphtheria of the pharynx and larynx with secondary ulcerative endocarditis From the pseudo-membranes as well as from the heart lesion he cultivated the streptococcus pyogenes.

D'Espine (10) has reported the isolation of a bacillus similar to that described by Loeffler, but the meagreness of his published data prevents a definite judgment as to the importance of his observations.

Early in 1888 v. Hoffmann (11) described the very frequent occurrence —in twenty-six out of forty-five cases—on the mucous membrane of the larynx, as well in diphtheria, scarlatina, measles, and simple catarrh as in quite normal conditions, of a bacillus apparently identical with the pseudo-diphtheritic bacillus of Loeffler. That is, it was very similar in morphology and growth to the diphtheria bacillus of Loeffler, but not virulent. A careful comparison of the genuine with the pseudo-diphtheritic bacillus led v. Hoffmann to the recognition of biological and morphological distinctions by which the two forms could be recognized. With this knowledge he again set to work, and from eight new cases of diphtheria he isolated the genuine Loeffler bacilli. But these, in one case, were not at all virulent for guinea-pigs, and in the others in a much less degree than those originally isolated by Loeffler. V. Hoffmann further isolated from three cases of measles the bacillus of Loeffler, the cultures in one case being virulent to guinea-pigs, in two not. Six times in nineteen cases of scarlatina he found Loeffler's bacilli, and these in no case were virulent. From the pharynx or larynx in eleven healthy cases he isolated Loeffler's bacillus four times; one of the cultures was virulent, the other not. V. Hoffmann seems to have made no animal experiments with the view of proving the etiological relation to diphtheria of these, Loeffler's bacilli, which he isolated. He simply tested their capacity of killing guinea-pigs by subcutaneous inoculations.

Recently, Roux and Yersin (17) have examined, partly by culture methods and partly morphologically, fifteen (?) cases of diphtheria, and state that they found the Klebs Loeffler bacilli in all. The record of their observations is so incomplete, however, that it does not appear whether or not they made cultures from all the cases. They apparently did not make use of the plate-culture method, but sowed the material from the pseudo-membranes directly upon tubes. Animal experiments were made, but were, so far as appears, comparatively few in number and apparently with cultures from but a few of the cases. These, in general, are said to be confirmatory of Loeffler's results. But as the local results of inoculations on injured mucous membranes are said by the authors, without detailed description, simply to be "false membranes," it is difficult to judge of their nature. The most suggestive part of their observations is the induction of symptoms of paralysis in animals by the injection of the fluids in which the bacilli had grown, and which had been freed from the germs by filtration. What bacteria, if any, other than those which they fixed upon as similar to the species isolated by Loeffler, developed in their cultures, does not appear, nor are such details of technique given as would inspire the reader with confidence that the cultures with which the animals were inoculated were actually pure cultures.

On the whole, the observations and experiments of these writers appear to be confirmatory of the work of Loeffler. But the records are so incomplete, and their technical procedures are of such a questionable nature and are so meagrely described, that it is very difficult to conclude whether the work is really of importance or not.

Since the perfection of the methods of staining bacteria in the tissues, a number of more or less careful simple morphological studies of diphtheritic lesions have been made, with the general result of showing what indeed had been demonstrated, even with the crude facilities of the older technique—that the diphtheritic membrane contains, as a rule, large numbers of bacteria of various shapes—the micrococcus form generally preponderating. This method also shows that the bacteria are sometimes present in the living tissue immediately about the seat of the false membrane, when this is formed, and that they sometimes are and sometimes are not to be found in the blood and internal organs.

If now we make a close summary of the actual results of the recent experimental work on diphtheria along the new lines of research, rejecting all simple morphological studies as incomplete and inconclusive, and confining ourselves to those cases only in which the culture methods have definitely revealed the identity of the bacteria, we find that from 16 cases of diphtheria out of 20 Loeffler isolated a bacillus which was capable of killing certain of the lower animals when introduced subcutaneously, and which, as tested from 6 cases when inoculated into a wounded mucous membrane, might induce an inflammation with a fibrinous pellicle. V. Hoffmann isolated the bacilli of Loeffler from 8 cases of diphtheria, but he cultivated them from 13 out of 33 cases of healthy and diseased air-passages in which there was no diphtheria present. Thus, while the Klebs-Loeffler bacilli have been cultivated 24 times from 28 cases thus examined, they have been found in 14 out of 34 cases which had nothing to do with diphtheria.

The results of animal inoculations with the pure cultures of Loeffler's bacillus are, on the whole, rather less convincing than the simple morphological and biological results. Certain animals are apt to die as the result of the inoculation of the Klebs-Loeffler bacillus, and inoculation on an injured mucous membrane may be followed by the formation of a fibrinous pellicle which somewhat, but not very closely, resembles the pseudo-membrane common in diphtheria. But if we recall how many agencies there are capable of producing a pseudo-membrane on the mucous surfaces, and how many forms of bacteria kill animals on subcutaneous injection with symptoms and lesions very similar indeed to those induced by Loeffler's bacillus, it seems to the writer that the reasons for identifying the animal with the human disease are neither definite nor convincing.

At present, then, it would seem that actual existing knowledge justifies us only in the conjecture that diphtheria is caused by some form or forms of bacteria. What the species is, or whether there are more than one, we do not know. We are confronted to-day with the same series of questions about which discussion has centred for many years. Are the diseases called pseudo-membranous pharyngitis and pseudo-membranous laryngitis or simple croup really, after all, identical with genuine diphtheria, differing only in that in one case there is a simple local lesion, while in the other there is a general systemic infection? It does not help us much in studying the etiology to assume that the systemic infection which stamps diphtheria as an acute infectious disease, and gives it clinically a more or less definitely isolated position, is the crucial test, because it is not only quite possible, but highly probable, that here, as for example in a poisoned wound of the finger, the process may in one set of cases be and remain local, while in another it may be local for a time and then become general, or be general almost from the outset.

That there is a form of croupous inflammation of the pharynx, larynx, and other air-passages, not due to active chemical irritants, and not occurring, as we say, "spontaneously," which is not obviously infectious and not apparently communicable from one individual to another, would seem to be well established by clinical observations. On the other hand, there is every reason to believe that many cases presenting the clinical features of simple croupous inflammation may be really diphtheria—as judged from the standpoint of their etiology—but in which, from some ineffective condition of the germs, from the smallness of the numbers inoculated, or from the resisting capacity of the individual, the action of the poison is limited to the seat of inoculation. We are constrained still to query whether the diphtheritic inflammation of the pharynx and other air-passages which so frequently accompanies scarlet fever and measles belongs to a different group from that which is not thus associated.

CHARACTER AND SCOPE OF OBSERVATIONS, TECHNIQUE, ETC.

The writer does not set before himself, in this paper, so ambitious a purpose as the complete solution of this problem of the etiology of diphtheria, which is one of the most complex and difficult, as it is one of the most important, questions in experimental pathology. But he wishes simply to record a series of observations made, in the light of modern science and with improved technical facilities, on a number of cases of diphtheria occurring in and near New York. When from one and another observer a sufficient number of such sets of cases shall have been studied and recorded, it is probable that then, and not until then, shall we be able to state definitely what the disease really is, the etiology of which we are seeking to discover. The problem is, at best, one of extraordinary difficulty, because we are liable to have on the exposed parts most often the seat of the local lesion of diphtheria the greatest variety of living bacteria, from the air, from water, and from food. These must all be carefully studied one by one, in each particular case, with all the precautions against error which experience and patience can furnish to the investigator.

The cases thus fully studied were those in which the clinical history left no room for doubt as to the diagnosis; all the fatal cases of extensive pseudo-membranous laryngitis, as well as pharyngitis, being regarded as diphtheria. They were most of them in children, were fatal, and early autopsies were made in nearly every case. The autopsies in most of the cases were made by a most competent and experienced observer—Dr. William P. Northrup—to whose zeal and courtesy I am indebted for much of the material. The parts to be subjected to examination in most of the cases were placed in carefully cleansed jars immediately on removal from the body, and brought or sent at once to the laboratory, where the detailed biological examination was begun without delay and carried on with the use of all the precautions against accidental contamination which the technical procedures formulated by Koch so admirably cover.

It has seemed to the writer wise, in undertaking these studies, not to seek to gather material for observation from a great variety of sources and from persons whose illnesses had run their course under the greatest variety of conditions, but to limit it largely to children, and especially to children which are placed under the general sanitary conditions prevalent in children's hospitals and asylums, where the cases may be closely observed from the outset and the surrounding conditions studied and more or less controlled.

Thus, while most of the cases came from one large foundling asylum,[1] in which the sanitary conditions are rather above the average in such institutions, in order to have some measure of comparison other cases were taken from other similar public institutions in other parts of New York City, and two cases occurred in private practice in one family in an adjacent town.

It will be readily appreciated, by those familiar with the details of such researches as these, that such a limitation of the problem is rendered imperative by the large expenditure of time which is required for the working out in full of any single case. Thus it was that a considerable amount of material from a number of cases of diphtheria (membranes, etc.), which was kindly placed by his colleague at the writer's disposal, was not made use of at all, partly because of the lack of time and partly because it was believed that a small group of cases fully worked out might lead to more reliable and tangible results than many only partially studied. In order, however, to avoid a misunderstanding of this selection of cases, it should be stated that material from every case of diphtheria which belonged in the group above indicated, and which came in proper condition for study into the writer's hands, was worked carefully through, and the results of all are recorded below.[2]

The primary task which the writer set before himself in the examination of these cases of diphtheria was, first, to ascertain, by the modern culture methods, what living microörganisms, if any, were present at and about the seat of the lesion or in the internal organs with sufficient frequency to justify the assumption that they might stand in an etiological relationship to the disease; to separate these in pure cultures, and study their life-history. Second, to learn by morphological examination of sections of the hardened and stained tissues what were the situation, distribution, and, so far as might be estimated, numbers of the microorganisms which might be present. Third, to ascertain the effects upon animals of the inoculation of the isolated species.

---

[1] These studies have extended over a period of two years, so that the cases from the large foundling asylum represent two distinct epidemics in successive years.

[2] I wish especially to acknowledge my indebtedness for material to Drs. J. Lewis Smith, R. Van Santvoord, L. Putzel, C. W. Oakes, and Mr. Charles Laylbach.

It seems hardly necessary to say that in all cases plate cultures were first made, and that these were manipulated in the usual way for the separation of individual species. Gelatine, agar, agar-glycerine, and blood-serum plates were used—the agar and agar-glycerine being the most generally efficient. Save for the gelatine cultures, the temperature of 37° C. (98.6° F.) was uniformly employed for the growth. The plate-cultures were mostly made either in shallow, covered dishes or in Esmarch tubes—generally in the former. Duplicate plates were in all cases made for the purpose of control, not only in the different cases, but for each part or organ subjected to examination. With this large number of plates, it was possible to make the sowings so dilute that the very large number of bacteria usually present in material from the air-passages could be readily obtained in isolated colonies.

It seemed unnecessary for the purpose in view, except in the first cases, to do more than isolate the similar species which developed in the different cases. Thus, a large number of forms which were familiar to the writer as occurring in the air, or water, or food, or those which control-cultures from the mouths and throats of healthy children, or those not suffering from diphtheria, showed to be common and unimportant, could be ignored after such observations on their characters as sufficed for their identification. Single colonies of different species developing on the plates were also for the most part ignored in the examination, since the probabilities were greatly against such isolated forms being of any significance. The task was thus narrowed down to the investigation of those forms of bacteria which developed in considerable numbers on the plates, and especially those similar forms which were found to occur on the plates made from different cases.

The colonies of these forms which were to be studied in detail were transferred first to a second set of plates and these, if evidently pure, were inoculated into tubes. In some cases, where large numbers of forms were present, several successive transfers from plate to plate were necessary to insure purity. The examination of the plates was made in the culture-room of the laboratory, which is kept scrupulously clean by the frequent washing of its shelves and floor with disinfecting solutions, and the air-supply to which, driven in by the ventilating fans, is filtered through frequently changed layers of dry cotton. In this way the air of the culture-room is so thoroughly free from germs that aërial contamination of the exposed plates during their examination was very infrequent. Thus it was possible to follow the development of colonies on the plates, even though they might be slowly growing forms, for long periods without danger of overgrowth by adventitious contaminations.

The writer had, fortunately, been able to acquire practical familiarity with the morphological and biological characters of Loeffler's bacillus from cultures which were the direct descendants of those isolated by Loeffler himself, and brought directly from Berlin.

Large numbers of duplicate sections from the hardened tissues of all the cases were prepared, and studied with a Zeiss $\frac{1}{12}$ homogeneous immersion lens and the Abbé condenser. Some of these sections were stained with Loeffler's alkaline methylene-blue solution, some by Gram's method, and some with the simple aqueous fuchsin solution. By the use of these three methods of staining in each case, the possibility of overlooking any form of bacteria in the tissues examined was largely reduced.

Having thus indicated the scope of the work and the precautions against error, it does not seem necessary to give an account of each case in detail, but to present the results in the form of a *résumé*.

The cases of diphtheria studied were twenty-four in number.[1]

CASES OF DIPHTHERIA EXAMINED.

CASE I.—*Clinical history:* Male, seven months. Feb. 8th, pertussis; 10th, diphtheria; 12th, intubation; 14th, death. *Autopsy*, twelve hours: Moderately thick and softening membrane in the trachea extending into the larger bronchi; tracheal wall at the lower end of tube eroded; broncho-pneumonia. *Bacterial examination—Morphological:* Enormous numbers of cocci, single, clustered, and in longer and shorter chains in all parts of the false membrane, particularly in its deeper layers; a few scattering, large, plump bacilli in the membrane. *Cultures: Streptococci*[2] in enormous numbers. Staphylococcus pyogenes aureus. A few scattering forms, the most abundant of which was a large bacillus resembling bacillus subtilis. In the false membrane in the trachea, the streptococcus exceeded the staphylococcus in numbers in about the proportion of 1300 to one, while in the bronchial exudate the proportion was about ten to one.

CASE II.—*Clinical history:* Male, two years and seven months. Jan. 28th, pertussis; 29th, acute tonsillitis, temperature 105°; 30th, diphtheria, albuminuria. Feb. 2d, broncho-pneumonia; 15th, death. *Autopsy*, three hours: Dense membrane in the lower part of trachea and in the larger and smaller bronchi; broncho-pneumonia in both lower lobes; parenchymatous nephritis. *Bacterial examination—Morphological:* In the bronchial exudate, especially in its deeper layers, were large numbers of cocci scattered in clumps and in longer and shorter chains. Similar cocci were found between the necrotic epithelial cells of the mucous membrane. A few chains of cocci were present in the lymph spaces of the mucosa and submucosa. A few stout bacilli were scattered through the false membrane. *Cultures: Streptococci* in large numbers. Staphylococcus pyogenes aureus. Aside from a few scattering colonies of variously shaped bacteria there were considerable numbers of a common white fluidifying yeast.

---

[1] The writer has made a large number of simple morphological examinations of sections of the false membranes and underlying parts from other cases of diphtheria than those now to be recorded; and, while it may be said of these that the cocci are, in almost every case, the prevailing form of bacteria, yet, as the morphological examination does not suffice to identify the species, the observations serve as contributing evidence of such moderate value, in the present state of our knowledge, that it does not seem worth while to do more than merely mention them here.

[2] For the sake of convenience, the morphological and biological character of this streptococcus will be given in detail below.

CASE III.—*Clinical history:* Umbilical phlegmon developed seven days after birth. At the same time pseudo-membrane was noticed on the pharynx, and there was considerable purulent conjunctivitis. With an increase in the amount of pseudo-membrane, and more intense phlegmonous inflammation about the umbilicus, the child died fourteen days after birth.[1] *Autopsy:* Dense firm pseudo-membrane over both tonsils and epiglottis; voluminous softening membrane in trachea and larger bronchi; broncho-pneumonia in both lower lobes with pulmonary congestion; parenchymatous degeneration of kidney; extensive umbilical phlegmon with suppuration. *Bacterial examination—Morphological:* Throughout the entire extent of the false membrane great numbers of cocci scattered and in chains. These were present as well in the deeper as in the superficial portions of the membrane. A small number were found scattered in the lymph spaces of the submucosa. A few scattering large bacilli were found in the membrane. *Cultures: Streptococci* in enormous numbers. Staphylococci pyogenes aureus and albus. A considerable number of large, long and plump, and short plump, round-ended bacilli. Cultures from the phlegmon showed both the streptococci and staphylococci.

CASE IV.—*Clinical history:* Female, three years and eight months. April 28th, varicella; 29th, croupy; 30th, dyspnœa and cyanosis, temperature 104.8°. May 2d, broncho-pneumonia, temperature 105.5°; 4th, death. *Autopsy*, twelve hours: Abundant pseudo membrane in pharynx, larynx, trachea, and larger bronchi, firm and closely adherent above, and softening in the trachea; broncho-pneumonia in both lower lobes; parenchymatous degeneration in kidneys. *Bacterial examination—Morphological:* Moderate numbers of cocci scattered and in longer and shorter chains in the false membrane. *Cultures: Streptococci* in enormous numbers and in nearly pure culture. A few staphylococci pyogenes aureus and a few other scattering forms on some of the plates.

CASE V.—*Clinical history:* Female, two years and eight months. May 5th, had been two weeks convalescent from scarlet fever when a pseudomembrane developed on the soft palate. Temperature 101°; albumin 80 per cent., granular casts. Two days later died, apparently from heart failure. *Autopsy*, six hours: Very slightly developed membrane, best formed in the trachea; moderate pulmonary congestion. *Bacterial examination—Morphological:* In the tracheal exudate a small number of cocci and a few stumpy bacilli. *Cultures: Streptococcus* colonies developed in small numbers on the plates, with a few colonies of staphylococcus pyogenes aureus and a few scattering forms.

CASE VI.—*Clinical history:* Female, three years. May 10th, pharynx red; 11th, scarlatina; 15th, pseudo-membrane in pharynx; 17th, diarrhœa; 20th, croupy cough, temperature 103°; 23d, weaker, dyspnœa; 25th, intubation, death. *Autopsy*, three hours: Firm pseudo-membrane

---

[1] This case was, in part, reported by Dr. J. Lewis Smith to the New York Pathological Society, June 13, 1888.

over both ulcerated tonsils. Pseudo-membrane in upper portions of trachea. Commencing broncho-pneumonia. Kidneys congested; parenchymatous degeneration. *Bacterial examination—Morphological:* The pseudo-membrane over the surface of the tonsils and the necrotic tissue on the tonsils near the ulcers and in the false membrane were crammed with cocci, single, massed, and in long and short chains. (See Plate I. and Plate II., Fig. 3.) The lymph nodules or follicles of the tonsils contained numerous single and chain cocci scattered between the cells. No bacteria were found by morphological examination in the kidney. *Cultures: Streptococci* were present in enormous numbers in the cultures from the pseudo-membrane of the tonsils and the necrotic tissue beneath it. They were also very abundant in the membrane from the upper portion of the trachea, less abundant in the softening portions below. Streptococci in pure culture and in considerable numbers from the kidney. Staphylococcus pyogenes aureus, most abundant in pharyngeal membrane.

CASE VII.—*Clinical history:* Male, two and a half years. May 24th, vomiting and croupy cough; 25th, pseudo-membrane in pharynx; 26th, temperature 102½°, croupy cough increasing, dyspnœa, death. *Autopsy,* thirty hours: Dense voluminous pseudo-membrane over both slightly roughened and ulcerated tonsils, over the entire larynx, and extending down to the larger bronchi; membrane softened below; commencing broncho-pneumonia. *Bacterial examination—Morphological:* In the pseudo-membrane over the tonsils, in the crypts of the tonsils, and in the necrotic tissue at the edges of the tonsillar ulcers were large numbers of cocci scattered and in short chains. A few stout bacilli in the superficial portions of the membrane. *Cultures: Streptococcus* colonies in enormous numbers in cultures from all parts of the membrane. Staphylococci pyogenes aureus and considerable number of scattering forms. Kidney cultures showed considerable number of streptococcus colonies, and nothing else.

CASE VIII.—*Clinical history:* Female, one year. Three days before death follicular stomatitis; during next night restlessness; then collapse and death. *Autopsy,* two and a half hours: Tonsils ulcerated and covered with firm pseudo-membrane; dense, firmly adherent membrane on the posterior pharynx, upper part of trachea, and œsophagus; entero-colitis; parenchymatous degeneration of kidney; commencing broncho-pneumonia. *Bacterial examination—Morphological:* Everywhere in the membrane and in the necrotic tissue near the ulcers of the tonsils were great numbers of cocci, scattered, clustered, and in large and short chains. These streptococci were less abundant in the tracheal membrane than above. Few scattering forms of bacilli. *Cultures: Streptococcus* colonies developed in vast numbers from the pseudo-membrane. Kidney cultures were negative, while from the spleen pure cultures of streptococci grew. Staphylococci pyogenes albus in small numbers grew from the membranes, while there were large numbers of the common "chain bacillus" of the mouth and a few scattering forms.

CASE IX.—*Clinical history:* Female, two years. May 27th, pneumonia, left side; temperature 106°. Seemed to recover. June 4th, pseudo-membrane on left tonsil. 5th, croupy cough, dyspnœa; intubation; broncho-pneumonia developed. 6th, death. *Autopsy,* twenty-one hours: Softening membrane over both tonsils; broncho-pneumonia, with pleurisy in left lower lobe. *Bacterial examination—Morphological:* In the membrane on tonsil considerable numbers of scattering cocci and streptococci; nothing else. *Cultures: Streptococcus* colonies grew in considerable numbers from the membrane; also large numbers of staphylococci pyogenes aureus, considerable numbers of fluorescent bacilli (doubtless from water) and of the "chain bacillus."

CASE X.—*Clinical history:* Male, three years. May 7th, scarlatina; 11th, pseudo-membrane on tonsils; 14th, otitis; 16th, swelling under left jaw. June 1st, abscess opened behind tonsil; 7th, death. *Autopsy,* four hours: Pseudo-membrane dense and firmly adherent at the base of tongue and over both tonsils, which were ulcerated. The membrane extended over the epiglottis, completely investing the larynx to below the vocal cords, where it became softened and shreddy. Chronic and commencing fresh broncho-pneumonia. Bronchial lymph glands greatly swollen and red. Kidneys, parenchymatous degeneration. *Bacterial examination—Morphological:* Everywhere in the pseudo-membrane and in the necrotic mucosa beneath were vast numbers of cocci, singly, clustered, and in long and short chains. A few isolated clusters of moderately stout, long bacilli were found near the surface of the pseudo-membrane. The lymph sinuses of the bronchial glands were distended, and contained an abundant network of fibrin. No bacteria were found in the spleen, kidney, liver, or bronchial lymph glands. *Cultures: Streptococci* in great numbers developed from various parts of the pseudo-membrane. The spleen cultures were negative, but a few colonies of streptococci, and nothing else, grew from the kidney and liver. Staphylococci pyogenes aureus grew in considerable numbers from the membrane, and a few from the liver. Some scattering forms of colonies developed from the membrane, the most abundant of which were the "chain bacillus" and a fluorescent water bacillus.

CASE XI.—*Clinical history:* Female, three years and two months. Feb. 1st, rectal abscess; 5th, croupy cough; 6th, intubation; 7th, death. *Autopsy,* sixteen hours: Pseudo-membrane in larynx, trachea, and larger bronchi. *Bacterial examination—Morphological:* Moderate numbers of cocci, mostly in pairs, in the pseudo-membrane. *Cultures: Streptococcus* colonies grew from the membrane in nearly pure culture. Staphylococci pyogenes aureus developed in small numbers, and a few scattering forms.

CASE XII.—*Clinical history:* Female, two and a half years. Feb. 9th, croupy cough, pseudo-membrane; 11th, intubation, broncho-pneumonia; 14th, temperature 102½°; 16th, death. *Autopsy,* two hours: Pseudo-membrane in pharynx, larynx, and trachea; ulcer in trachea at end of tube; broncho-pneumonia in both lower lobes. *Bacterial examination—Morphological:* Large numbers of cocci, many in chains, in the pseudo-membrane; few scattered bacilli. *Cultures: Streptococcus* colonies in considerable numbers grew from the membrane. Also staphylococcus pyogenes aureus and a few scattering colonies of stout, large bacilli.

CASE XIII.—*Clinical history:* Female, two years and two months. Brought to the hospital with vulvitis, which developed into erysipelas about vulva. Eight days later, false membrane appeared on the tongue; then croup followed, with death in five days. *Autopsy,* two hours: Vulva denuded of epithelium, dusky red. Dense, firm, closely adherent membrane on velum and uvula; tonsils ulcerated and covered with false membrane, which extended down over the posterior surface of the epiglottis and onward to just below the vocal cords; no broncho-pneumonia. *Bacterial examination—Morphological:* Large numbers of cocci, single, in pairs and chains, in the deeper layers of pseudo-membrane, and extending deeply into the underlying tissue, accompanied here, when in large numbers, by necrosis. Other scattering forms were abundant. Scattered clusters of slender bacilli were found in the pseudo-membrane near the vocal cords, especially in the most superficial portions. *Cultures:* In the pseudo-membrane from the tonsils, pharynx, and larynx, enormous numbers of *streptococci* in nearly pure cultures; a few staphylococci pyogenes aureus and a few scattering forms, mostly large bacilli.

CASE XIV.—*Clinical history:* Female, two months. Returned to hospital from out-nursing with bad cough, and after being for eight days in room with considerable number of children, sick and well, developed diphtheria, rapidly followed by broncho-pneumonia, with death in two days. *Autopsy,* eleven hours: Pseudo-membrane over both tonsils; on posterior pharynx and in larynx and trachea down to bifurcation membrane softening. *Bacterial examination—Morphological:* The pseudo-membrane had in many places considerably involved the superficial portions of the mucosa. Moderate numbers of cocci and streptococci in the pseudo-membrane, together with a considerable number of stout, long bacilli in the more superficial portions. *Cultures:* False membrane on tonsils, pharynx, and larynx all alike showed large numbers of *streptococci.* There were also many staphylococci pyogenes aureus and a considerable number of colonies of coarse, stumpy bacilli.

CASE XV.—*Clinical history:* Male, three years. Returned to hospital Dec. 3d. On Dec. 13th developed diphtheria; 20th, intubation; 23d, broncho-pneumonia, cyanosis, death. *Autopsy,* six hours: Tonsils swollen, not ulcerated. Small patch of pseudo-membrane on pharynx, and extending from the tip of the epiglottis to the medium bronchi, firm and closely adherent above. Congestion and commencing broncho-pneumonia in both posterior lower lobes. Cortex of kidney thick and opaque, spleen and liver appear normal. *Bacterial examination—Morphological:* Surfaces of tonsils and crypts intact, with small masses of diplococci and streptococci here and there in the crypts. In the pseudo-membrane enormous numbers of cocci in pairs and chains, especially in the deeper layers and in the necrotic tissue of the submucosa beneath. Other scattering forms were present, but none abundant. No bacteria found in the viscera. *Cultures:* Large numbers of *streptococcus* colonies developed from scrapings of tonsil crypts, from various parts of the pseudo-membrane, and from the lungs. Cultures of liver, kidney, and spleen were negative.

CASE XVI.—*Clinical history:* Male, three and a half years. Six months in hospital. Dec. 20th, croupy cough, relieved by emetic. 21st, croup symptoms continued; no pseudo-membrane seen on pharynx. 22d, died. *Autopsy,* four hours: Tonsils swollen with large pit-like crypts. Dense, firm pseudo-membrane extends from tip of epiglottis down to bifurcation of trachea; firmly attached as far down as vocal cords, below this loose. Bronchitis, but no broncho-pneumonia. Bronchial and tracheal lymph glands swollen and red. Kidneys, parenchymatous degeneration. Ulcerative gastritis and ileo-colitis. *Bacterial examination—Morphological:* In the pseudo-membrane very few scattered bacteria were found. There were a few cocci, mostly on the surface, and a few short, stout bacilli. There was no necrosis of the tonsils, and but few bacteria in the crypts. No bacteria of any form were found in either bronchial or lymph glands, or in the kidney, liver, or spleen. *Cultures:* In the pseudo-membrane and tonsil crypts were found a considerable number of short, stout, round-end bacilli and several scattering forms. No streptococcus colonies developed. Cultures from the lymph glands and viscera were sterile.

CASE XVII.—*Clinical history* is wanting, save that the child, with several others in the same hospital, was suffering from measles when the diphtheria developed. No pseudo-membrane was seen in the pharynx, and the child died soon after the diphtheritic symptoms manifested themselves. *Autopsy:* Tonsils much swollen, with somewhat ragged surfaces. Voluminous, firm pseudo-membrane from the tip of epiglottis down to medium bronchi, loosened and softening below. Commencing broncho-pneumonia. Cortex of kidney slightly thickened, liver and spleen appeared normal. *Bacterial examination—Morphological:* Large numbers of cocci, streptococci, and diplococci in tonsil crypts and over the eroded surfaces. Similar forms in moderate numbers in the deeper layers of the pseudo-membrane. Aside from these, only scattering forms. No bacteria found in kidney, liver, or spleen. *Cultures:* Large numbers of *streptococci* from all parts of the pseudo-membrane and from the tonsil crypts, also some scattering forms. A few streptococcus colonies developed from the kidney and spleen; liver cultures were negative.

CASE XVIII.—*Clinical history:* Child, one year. Had been in hospital for six months with bronchitis. Measles developed on Dec. 24th, and ran a course of ordinary severity; croupy symptoms began early in the disease, and on Jan. 8th diphtheritic symptoms were pronounced, though there was no pseudo-membrane on the pharynx. The child died on Jan. 9th. *Autopsy:* No pseudo-membrane on pharynx, but on the larynx below the epiglottis were numerous, small, irregular, firm, closely adherent patches of pseudo-membrane; the tracheal and bronchial glands were red and swollen. Viscera not examined. *Bacterial examination—Morphological:* Large numbers of cocci, single, paired, and in chains, were irregularly grouped, especially in the deeper layers of the pseudo-membrane and sparsely infiltrated the mucosa beneath. No bacteria were found in the tracheal and bronchial glands. *Cultures:* Small numbers of various scattering forms, among which a few staphylococci pyogenes aureus were present. The *streptococcus* was present in large numbers.

CASE XIX.—*Clinical history:* Child, one year and eight months. Four months in hospital with diarrhœa. The child was attacked with measles of a severe form, with high temperature, vomiting, and cough. Croupy symptoms appeared on the third day, and increased steadily till the sixth, when tracheotomy was performed. Slight improvement followed, but the child died thirty-six hours after the operation. *Autopsy:* Small particles of pseudo-membrane on tonsils. Dense voluminous membrane in larynx and trachea. Cheesy bronchial glands, with localized tubercular inflammation of the lungs, and fresh broncho-pneumonia. *Bacterial examination—Morphological:* Pseudo-membrane closely adherent in places with much involvement of the epithelium, and superficial necrosis of the submucosa. Enormous numbers of cocci, single, in pairs and chains, in the deeper layers of the pseudo-membrane, in the necrotic mucosa, and infiltrating in smaller numbers the lymph spaces of the submucosa (see Plate III.). No bacteria found in the lymph glands. *Cultures:* From the pseudo-membrane of the larynx and trachea were cultivated large numbers of *streptococci.* Besides these a few colonies of staphylococci pyogenes aureus and albus, and a few scattering forms.

CASE XX.—*Clinical history:* Child, two years. Twenty-three days in hospital with diarrhœa. Measles developed, running a severe course with high temperature. A croupy cough developed early with dyspnœa and symptoms of pneumonia. On the eighth day intubation was practised without benefit, and the child died in a few hours. *Autopsy:* Tonsils were swollen; the mouth of the crypts dilated. No false membrane was found, but both laryngeal and pharyngeal mucous membranes were swollen and intensely red. The bronchial glands were red and swollen. The lungs were congested. *Bacterial examination—Morphological:* The epithelium covering the surfaces of the tonsils and lining the crypts was for the most part intact, but here and there in the depth of the crypts small patches of necrotic epithelium were found, which were infiltrated with streptococci. A few scattering cocci and streptococci were found on the surface of the intact epithelium in the crypts. In the ventricles of the larynx and about the rima glottidis were considerable numbers of cocci and streptococci, with a few stout bacilli. The epithelium in the ventricles of the larynx was in places partially detached and necrotic, and in these regions numerous streptococci were found beneath the epithelium, and in the superficial layers of the submucosa. In this case, then, in which there was no pseudo-membrane, in two places, at least, there appeared to be a commencing invasion of the tissues with streptococci. No bacteria were found in the lymph glands. The air-spaces of the lungs contained enormous numbers of streptococci and cocci. *Cultures:* Scrapings from the tonsil crypts and from the region of the vocal cords showed considerable numbers of *streptococci,* together with scattering forms.

CASE XXI.—*Clinical history:* Child, one year and six months. In hospital six months. Measles running a severe course, with temperature 104°. Dyspnœa was developed early, and the child died twenty-four hours after the commencement of the disease. *Autopsy:* Tonsils were swollen and ragged. In the pharynx, at the base of the epiglottis, on either side, was a small, irregular patch of pseudo-membrane, dense and closely adherent. Small white-surfaced ulcers were found just below the rima glottidis. No pseudo-membrane visible elsewhere, but the larynx was of a dusky red color. *Bacterial examination—Morphological:* The bloodvessels of the mucous membrane were widely dilated. In the small patches of pseudo-membrane in the pharynx, as well as at the seat of the ulcer, below the rima glottidis, there were large collections of cocci, mostly in short chains, which penetrated the submucosa, and on either side extended beneath the intact epithelium. In the vicinity of these regions, but especially in the pharynx, there were considerable collections of cocci and streptococci lying closely packed against the surface, or infiltrating the superficial epithelial layers. In the regions where these bacteria were numerous the epithelium was either necrotic or was much thickened and peeling off in large masses. The tonsils were intact, but in the mouths of the crypts, as well as entangled in the mucus of the larynx, were a few cocci, single or in short chains, and a few scattering forms of other bacteria. In this case we seem to have the commencement of the formation of the pseudo-membrane, and the invasion of the adjacent tissues with streptococci. *Cultures:* A moderate number of *streptococci* grew from the scrapings of the pseudo-membranous patches, together with considerable numbers of scattering forms. Among the latter were a few staphylococci pyogenes aureus.

CASE XXII.—*Clinical history:* Child, six days old. Developed severe umbilical phlegmon. Three days after this, diphtheria appeared, with severe local and general symptoms, with death on the following day. *Autopsy,* twenty-six hours: A dense, thick, firmly adherent membrane on the posterior surface of the epiglottis, extending down over the vocal cords and into the larger bronchi; commencing broncho-pneumonia, with much pulmonary congestion; parenchymatous degeneration in kidney. *Bacterial examination—Morphological:* In the false membrane enormous numbers of cocci, mostly single and in pairs, but many were in chains and irregular clusters. Many similar cocci were present in the lymph spaces of the submucosa and in the mucous glands of the trachea and larger bronchi. *Cultures: Streptococci* in large numbers. Staphylococcus pyogenes aureus. A few colonies of large bacilli resembling bacillus subtilis. Cultures from the tissues about the umbilical phlegmon revealed large numbers of streptococci and staphylococcus pyogenes aureus.

CASE XXIII.—*Clinical history:* Female, six years. Taken sick on Monday, with croupy cough and dyspnœa. No pseudo-membrane was visible. There was no evidence of systemic infection, but the dyspnœa increasing, tracheotomy was advised and refused by the parents. On Monday the child died. *Autopsy:* No pseudo-membrane in the pharynx. A dense, voluminous pseudo-membrane extended from the base of the epiglottis to the bifurcation of the trachea; this was firm and closely adherent above, leaving raw, rough surfaces when pulled away. Microscopical examination showed that, especially above, the pseudo-membrane had involved the external layers of mucous membrane *Bacterial examination—Morphological:* Few scattering bacteria were present in the pseudo-membrane, mostly on the surface; these were mostly larger and smaller short, stout bacilli. *Cultures:* No streptococci and no staphylococci were found. There were two forms of bacilli in considerable abundance, one a form common in the healthy mouth, and the other a short, plump, round-end bacillus. There were three or four scattering forms, mostly bacilli.

CASE XXIV.—*Clinical history:* Female, two and a half years. Croupy cough, dyspnœa, no membrane in pharynx, but strips of membrane coughed up. Constitutional symptoms became graver; tracheotomy; symptoms of broncho-pneumonia; death on the sixteenth day. *Autopsy,* twenty-four hours: Voluminous membrane in larynx, and trachea softened and loose below; no membrane in pharynx. Commencing broncho-pneumonia. Kidney swollen, cortex white and thick. Liver and spleen normal in appearance. *Bacterial examination—Morphological:* Pseudo-membrane thickly permeated by cocci, diplococci, and streptococci, with a few other scattering forms. Streptococci in considerable numbers in the lymph spaces of the submucosa. A few in the ducts and acini of the tracheal mucous glands and in the tracheal lymph nodes; large numbers of streptococci in the tissues about the tracheotomy wound; no bacteria of any form found in the kidney, liver, or spleen. *Cultures: Streptococci* in enormous numbers in the membrane with few scattering forms. Pure cultures in large numbers of streptococci in spleen, lungs, and liver. (Kidney cultures lost by accident.)

Before proceeding to study the characters and life-history of the streptococcus, to the importance of which our studies thus far seem to point, we will group the cases of diphtheria examined and briefly summarize the results.

(To be concluded.)

ON THE USE OF HYPNOTICS, SEDATIVES, AND MOTOR DEPRESSANTS IN THE TREATMENT OF MENTAL DISEASES.

BY T. S. CLOUSTON, M.D.,
PHYSICIAN-SUPERINTENDENT, ROYAL EDINBURGH ASYLUM; LECTURER ON MENTAL DISEASES, EDINBURGH UNIVERSITY; PRESIDENT OF THE MEDICO-PSYCHOLOGICAL ASSOCIATION.

FOUR cases out of five of recent mental disease have either sleeplessness, or active brain excitement, or morbid motor activity, as part of their symptoms at some time. The other symptoms present, mental and bodily, often seem to be of less importance than those to the patient's relatives and to his physician. Their urgency and troublesomeness seem to call for direct and immediate medical treatment in a very large number of such cases. While as physicians we fully recognize that these are symptoms, and not the disease itself, yet they all so manifestly tend toward brain exhaustion, that it is very natural to adopt means for their relief. And the most obvious medical means are the use of hypnotic, sedative, and motor depressant drugs. I do not say that such drugs are as curative as they seem, though I fully admit that their use is sometimes curative. And few practitioners but frequently find their use inevitable. The temptation to use them is sometimes overwhelming. But the dangers of using them to excess are great and numerous. I hope I am not wrong in the opinion I have formed that as they have been used they have often done more harm than good as regards cure, though it may be said that the sum total of the present relief they have afforded has been so great a boon to the sufferings of humanity, that their disuse would be a cruelty not to be thought of.

To go into their use fully in each form and phase of mental disease, in each several temperament, diathesis, and age would require a treatise, and our knowledge is not yet exact enough to enable anyone to do so. Even fully to state the principles that should guide us in their use, so far as I know them, and the risks to be guarded against would take up much time. Now-a-days most of us want to take our medical reading in a concentrated form. I have had much experience of their use and some proofs of their abuse. I have experimented carefully with many sedative and hypnotic drugs, but I find it very difficult to condense my experience, and lay down rules or principles of general application that would be of use to others.